Cebpa Cas9-CKO Strategy

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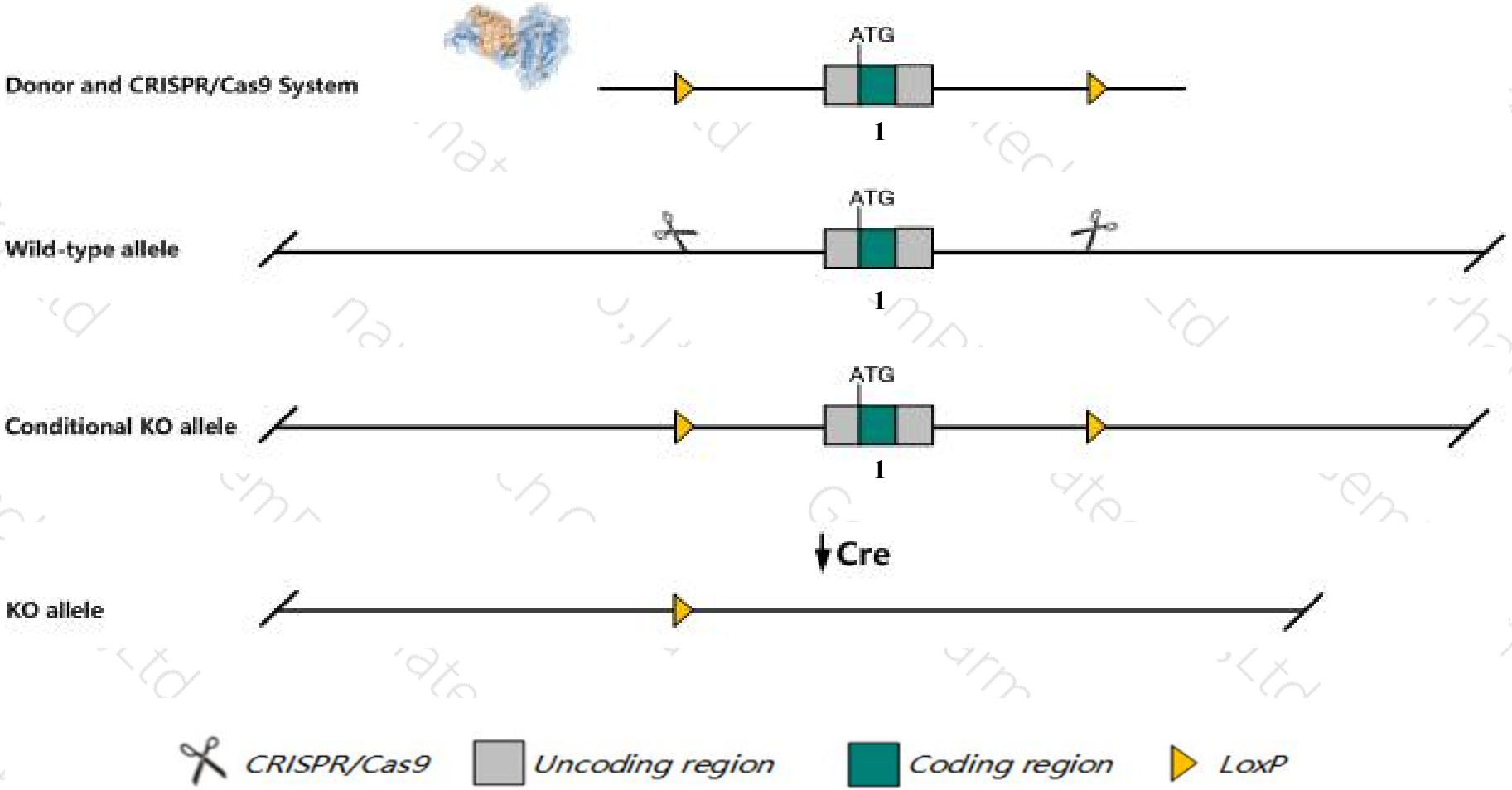
Project Overview

Project Name	***Cebpa***
Project type	**Cas9-CKO**
Strain background	**C57BL/6JGpt**

Conditional Knockout strategy

This model will use CRISPR/Cas9 technology to edit the *Cebpa* gene. The schematic diagram is as follows:



Technical routes

- The *Cebpa* gene has 2 transcripts. According to the structure of *Cebpa* gene, exon1 of *Cebpa-201* (ENSMUST00000042985.10) transcript is recommended as the knockout region.The region contains all of the coding sequence.Knock out the region will result in disruption of protein function.

- In this project we use CRISPR/Cas9 technology to modify *Cebpa* gene. The brief process is as follows:CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.

- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

Notice

- According to the existing MGI data,homozygotes for targeted null mutations exhibit defects of the liver, neutrophils, lung, and brown fat, resulting in impaired glycogen storage and lipid accumulation, hypoglycemia, reduced uncoupling protein, and neonatal lethality.
- The *Cebpa* gene is located on the Chr7. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This strategy is designed based on genetic information in existing databases.Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

Gene information （NCBI）

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Cebpa CCAAT/enhancer binding protein (C/EBP), alpha [Mus musculus (house mouse)]

Gene ID: 12606, updated on 22-Mar-2020

Summary

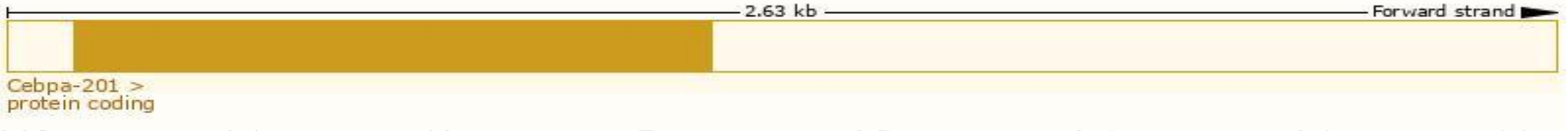
Official Symbol	Cebpa provided by MGI
Official Full Name	CCAAT/enhancer binding protein (C/EBP), alpha provided by MGI
Primary source	MGI:MGI:99480
See related	Ensembl:ENSMUSG00000034957
Gene type	protein coding
RefSeq status	REVIEWED
Organism	Mus musculus
Lineage	Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha; Muroidea; Muridae; Murinae; Mus; Mus
Also known as	C/ebpalpha, CBF-A, Cebp
Summary	This intronless gene encodes a transcription factor that contains a basic leucine zipper (bZIP) domain and recognizes the CCAAT motif in the promoters of target genes. The encoded protein functions in homodimers and also heterodimers with CCAAT/enhancer-binding proteins beta and gamma. Activity of this protein can modulate the expression of genes involved in cell cycle regulation as well as in body weight homeostasis. The use of alternative in-frame non-AUG (CUG) and AUG start codons results in several protein isoforms with different lengths. Differential translation initiation is mediated by an out-of-frame, upstream open reading frame which is located between the CUG and the first AUG start codons. [provided by RefSeq, Sep 2014]
Orthologs	human all

Transcript information（Ensembl）

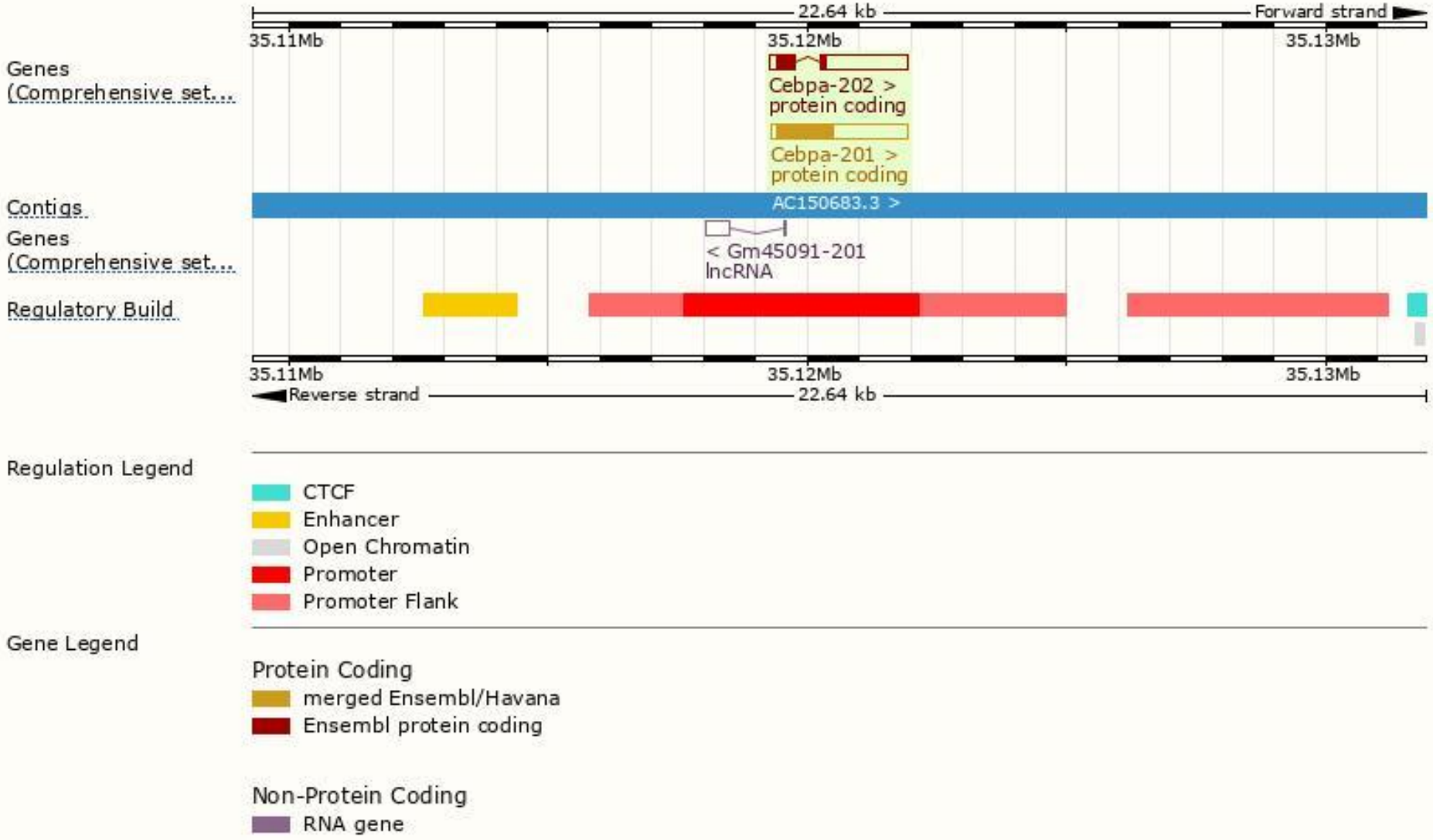
The gene has 2 transcripts,all transcripts are shown below:

Name	Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
Cebpa-201	ENSMUST00000042985.10	2626	359aa	Protein coding	CCDS21145	P53566	TSL:NA GENCODE basic APPRIS is a system to annotate alternatively spliced transcripts based on a range of computational methods to identify the most functionally important transcript(s) of a gene. APPRIS P1
Cebpa-202	ENSMUST00000205391.1	2161	159aa	Protein coding	-	A0A0U1RPE0	TSL:5 GENCODE basic

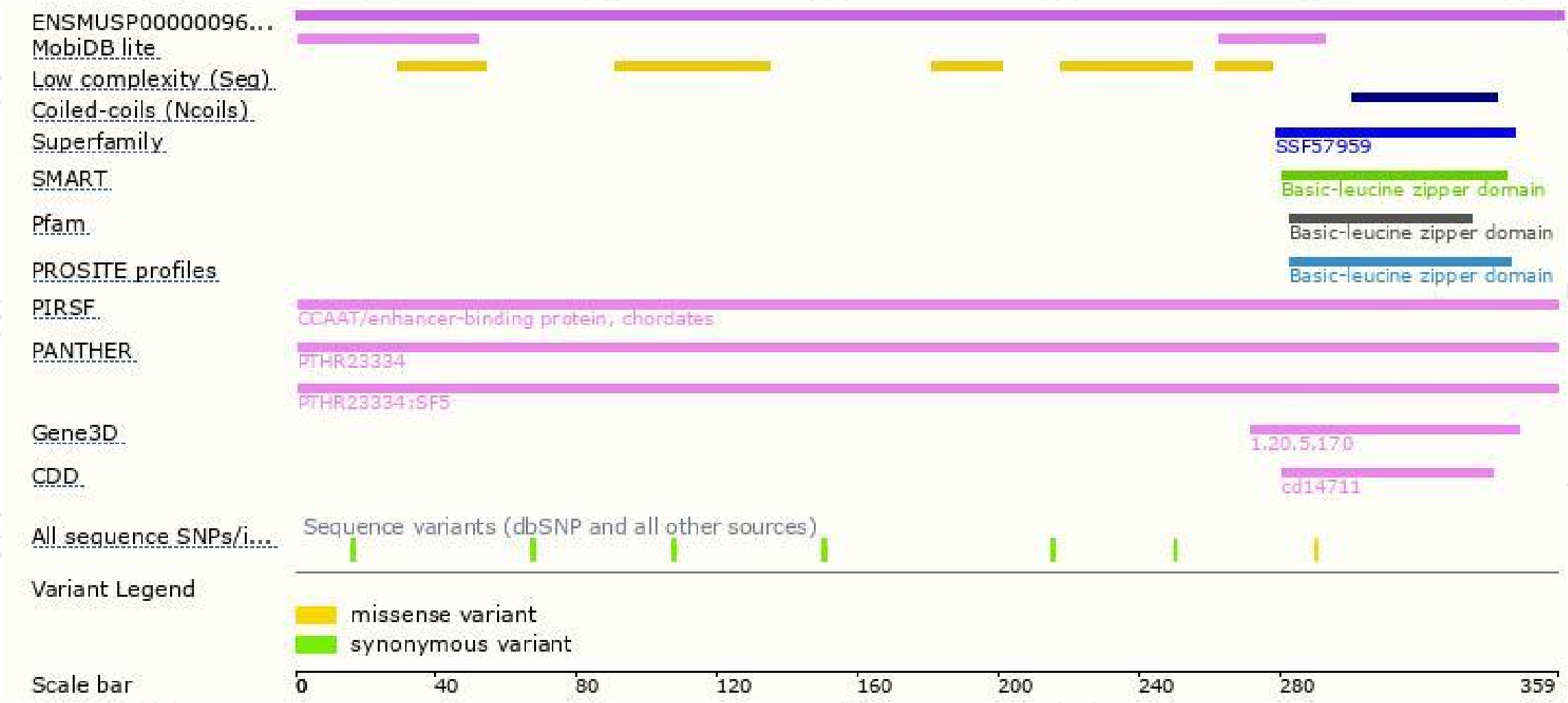
The strategy is based on the design of *Cebpa-201* transcript,the transcription is shown below:



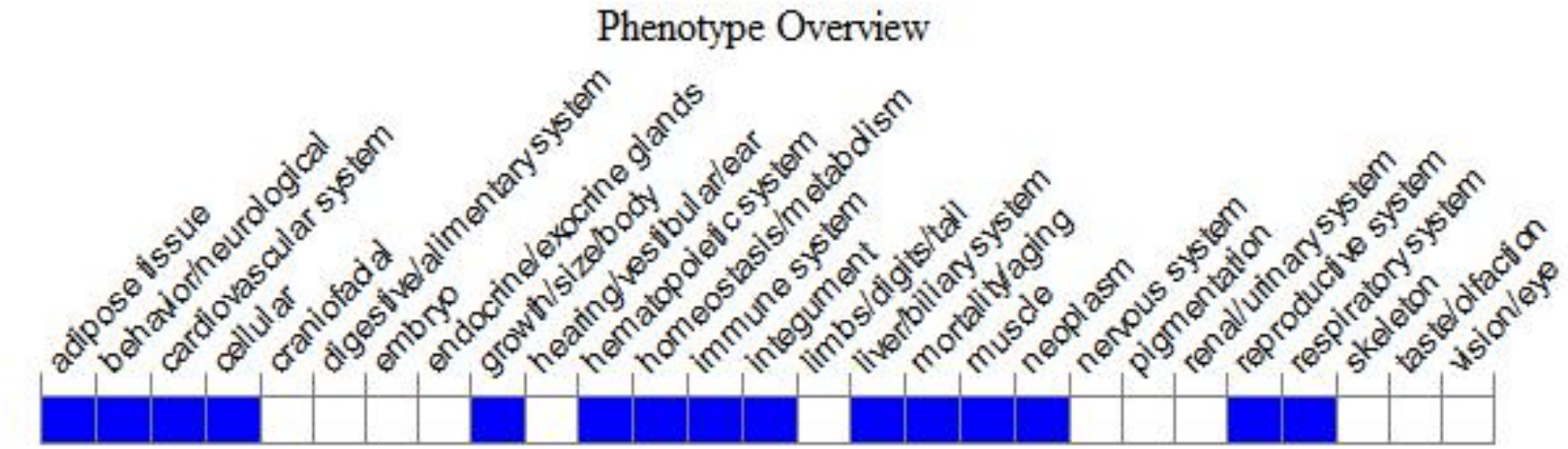
Genomic location distribution



Protein domain



Mouse phenotype description(MGI)



Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

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If you have any questions, you are welcome to inquire.

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